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Prevalence and Associated Risk Factors of *Neisseria gonorrhoeae*, *Treponema pallidum*, *Trichomonas vaginalis* and *Candida* species Infections among Symptomatic Patients Attending at Gondar Town Hospitals and Health Centers, Northwest Ethiopia

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ABBREVIATIONS

AIDS	Acquired Immunodeficiency Syndrome
ATCC	American Type Culture Collection
CDC	Centers for Disease Control and Prevention
CLSI	Clinical and Laboratory Standards Institute
HIV	Human Immunodeficiency Virus
ICDC	Intracellular Diplococcic
PMNL	Polymorphonuclear Leukocyte
RPR	Rapid Plasma Reagin
SPSS	Statistical Package for Social Sciences
STI	Sexually Transmitted Infections
TMT	Thayer Martin Media
WHO	World Health Organization

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ABSTRACT

Background: Sexually transmitted infection (STI) is a major global cause of acute illness, infertility, long-term disability and death, with serious medical and psychological consequences to millions people. In Ethiopia epidemiological study on STI is limited.

Objective: The aim of this study was to determine prevalence and associated risk factors of *Neisseria gonorrhoeae*, *Treponema palladium*, *Trichomonas Vaginalis* and *Candida species* infections.

Methods: A cross sectional study was conducted among STI patients attending Gondar town hospitals and health centers between April and August 2013. One hundred twenty consecutive symptomatic study participants were enrolled in the study. Socio-demographic data from each study participant was collected using a semi-structured questionnaire. Samples (blood and urogenital discharges) were collected and laboratory diagnosis for STI was done. Antimicrobial susceptibility test of *N. gonorrhoeae* was done using disc diffusion technique. Data were analyzed with descriptive statistics and logistic regression using SPSS version 20.

Results: The overall prevalence of laboratory confirmed STI was 89(74.1%). *Candida species*, *T. palladium*, *N. gonorrhoeae* and *T. vaginalis* infections were accounted 39(32.5%), 36(30%), 25(20.8%) and 17(14.2%), respectively. The prevalence of *Candida species* and *T. palladium* were higher in HIV positive patients. Unmarried patients were at higher risk of *T. vaginalis* infection (AOR 5.9, 95%CI 1.18-29.9, p=0.036) as compared to married study subjects. HIV infection (AOR 11.9, 95% CI 3.0-46.4, p=0.00) and regular alcohol intake (OR 3.9, 95% CI 1.2-12.9, p=0.02) were also significant predictors of *T. pallidum* infection. Isolated *N. gonorrhea* was resistant to tetracycline (100%), penicillin (76%), ciprofloxacin (52%) and ceftriaxone (48%) and 80% of the isolates showed multidrug resistance.

Conclusions: Higher prevalence of *N. gonorrhoeae*, *T. pallidum*, *T. vaginalis*, and *Candida species* infections among symptomatic STI suspected patients were observed. Being HIV positive and regular alcohol intake were significant predictors of STI infections. Drug resistance of *N. gonorrhoeae* to ciprofloxacin and ceftriaxone were very high. Thus, treatment of gonococcal infection should be supported with drug susceptibility testing.

Key words: *Candida species*, *N. gonorrhoeae*, STI, sexually transmitted infection, *T. pallidum* *T. vaginalis*

1. INTRODUCTION

Sexually transmitted infections (STIs) are group of infections or communicable diseases in which the primary mode of transmission is through sexual contact. Some of the common STIs include: bacterial vaginosis, herpes, Chlamydia, trichomoniasis, gonorrhea, Hepatitis B virus, Human Immunodeficiency Virus (HIV)/ Acquired immunodeficiency syndrome (AIDS) and syphilis (1). STI is classified according to the type of organism causing the infection, which can be bacterial, fungal, viral or of parasitic origin. *Neisseria gonorrhoeae* (*N. gonorrhoeae*), *Chlamydia trachomatis*, *Gardnerella vaginalis*, *Treponema palladium* (*T. pallidum*), *Trichomonas vaginalis* (*T. vaginalis*) are some of the most incriminated as etiological agents of STI (2).

Neisseria gonorrhoeae, the causative agent of gonorrhea, is an intracellular human pathogen that primarily colonizes the urogenital tract. Gonorrhea remains a global public health problem. Nowadays worryingly, *N. gonorrhoeae* has developed high-level resistance to all traditional antimicrobials used for the treatment of gonorrhea, like penicillin, tetracycline and fluoroquinolone (3). *Trichomonas vaginalis*, a small, motile, flagellated, protozoan parasite, causes trichomonal vaginitis (4). *Trichomonas vaginalis* is opportunistic pathogen that could grow abnormally when the vagina ecosystem or the urethra in male has a distorted environmental balance (2). *Treponema pallidum*, a thin flagellated spirochete, is the etiologic agent of syphilis. Syphilis is typically acquired via sexual contact but the disease can also be transmitted transplacentally and by exposure to blood or lesion exudates from infected persons in the primary and secondary stages of the disease (5).

Sexually transmitted infections are a major global cause of acute illness, infertility, long-term disability and death, with serious medical and psychological consequences to millions of men, women and infants (6). Sexually transmitted infections are a major public health problem worldwide. According to 2005 the world health organization (WHO) report, more than 448 million new cases of curable STI (Chlamydia, gonorrhea, trichomoniasis and candidiasis) and a total of 347 million new cases of gonorrhea, trichomoniasis and syphilis occur every year worldwide in people aged between 15 and 49 years old. Among this majority of cases were in developing countries. For instance, over 22% of adults with *N. gonorrhoeae* were found in the

WHO African region (7). In the developing world, STI present major health, social and economic problems, leading to considerable morbidity, mortality and stigma. The prevalence rates apparently are far higher in developing countries where STI treatment is less accessible (8).

In sub-Saharan Africa 67% of infertility is due to STI associated tubal blockage. Post-infection tubal damage is responsible for 30 - 40% of cases of female infertility which lead stigmatization and divorce. In pregnant women with untreated early syphilis and gonococcal infection results 40% and 10% perinatal mortality respectively. Worldwide, 1000 - 4000 newborn babies become blind every year because of eye infection (9). Like other developing countries, in Ethiopia, the burden of STI is high but there is little information on the incidence and prevalence of STIs. This is due to that people with STIs who have minor or no symptoms do not seek treatment at public health facilities. They usually tend to take self-prescribed drugs or go to private pharmacies to buy treatment without consulting trained health workers, lack of information on STIs, stigma associated with attending public STI clinics, the formal public health facilities also do not report all STI cases properly and comprehensively and economic factors (10).

Studies indicated that some infections or co-infections of STI have the chance of increasing the risk of HIV transmission. STIs associated with *T. vaginalis*, *N. gonorrhoeae*, *T. pallidum*, and *C. albicans* infections enhance the acquisition and transmission of HIV (11- 13).

In resource limited countries the absence of etiologic diagnostic capacity has forced health care providers to rely on a syndrome based approach to STI management, yet many STIs have common symptoms or are asymptomatic and therefore go undetected and untreated (6). Improvement in the management of STIs can reduce the incidence of HIV infection in the general population by about 40%. Prevention and treatment of STIs are therefore critical components of HIV prevention and treatment strategies (9). Identifying the etiology and prevalence of STI causing organisms is important to deliver appropriate treatment and decrease the risk for HIV transmission. Therefore, this study primarily aimed to determine the prevalence and associated factors of *N. gonorrhoeae*, *T. vaginalis*, *T. pallidum* and *Candida species* among symptomatic STI suspected patients attending Gondar town hospitals and health centers.

2. LITERATURE REVIEW

Sexually transmitted infections constitute a major threat to health worldwide particularly in developing countries in which the prevalence is much higher. STI and their complications are amongst the top five diseases in which adults seek help in outpatient clinics (14).

A study conducted in California STI clinic among homosexual males indicated that the prevalence of gonorrhea in different anatomic sites were 6.9% rectal, 6.0% urethral, and 9.2% pharyngeal (15). On the other hand, a study from Colombia showed that there was 12.3% prevalence of STI with the most frequent etiology was candidiasis (11%), *N. gonorrhoeae* (1.4%) and *T. vaginalis* and *T. pallidum* (0.8% each) (1). Moreover, the study conducted in Marry land STI clinic to compare culture methods indicated that the prevalence of *N. gonorrhoeae* was 29% of genital cultures, 3% of pharyngeal cultures, and 29% of rectal cultures (16).

In Mongolia among female patients the prevalence of *N. gonorrhoeae* and *T. vaginalis* were 11% and 8% respectively (17). On the other hand in Francisco gays STI clinic there were prevalence of 0.6% for *T. vaginalis* and 11.4% for *N. gonorrhoeae* (18). Furthermore, study among men patients indicated that the prevalence of *N. gonorrhoeae* and *T. vaginalis* were 5.1% and 2.8% respectively. The prevalence of *T. vaginalis* was higher in the age groups of 30 years or older patients. The presence of urethral discharge and nongonococcal urethritis were independent predictors of *T. vaginalis* infection (19).

A study conducted in India, to determine prevalence of gonorrhea & its co-infection with other ulcerative, non-ulcerative STI & HIV infection shows there was 14.4% prevalence of gonorrhea. Gonorrhoeae was the most common co-infection with syphilis (6.3%) followed by *C. albicans* (2.5%) and HIV (2.2%). The prevalence of syphilis among ulcerative STI patients was 36.9% while 1.8% *T. vaginalis* infection was observed among non-ulcerative STI patient (20). According to 1999 WHO estimate sub-Saharan Africa was the second to have largest number of new infections of curable STI which accounts 69 million new cases. But, the highest rate of new cases per 1000 population has occurred in sub-Saharan Africa (3). In Nigeria, the prevalence of STI was 55% of which the prevalence of *C. albicans* and *T. vaginalis* were 1.5% and 27% respectively. Age, sex and marital status were significantly associated with STI (21). Moreover,

patients who had more than one sexual partner in the past 3 months, having a history of a bacterial STI, being black Caribbean ethnicity, and living in a deprived area were significant predictors of STI (22). Beside this, another study in Nigeria indicated that the prevalence of *N. gonorrhoeae* was 25% and none of the study participants were infected with *T. vaginalis* (23).

Apart from being serious health, social and economic problems, STIs predispose to HIV infection (24). The presence of an untreated STI can increase the risk of both acquisition and transmission of HIV by a factor of up to 10 (7). Study conducted on HIV infected women, Washington, indicated that there were prevalence of 11%, 6% and 4% of *N. gonorrhoeae*, *syphilis* and *T. vaginalis* respectively (25). A comparative study on the prevalence of STI pathogens in HIV infected and non-infected women from Nigeria indicated that higher prevalence of *C. albicans* (77%) and *T. vaginalis* (12%) among HIV-positive patients were observed as compared to the prevalence of *C. albicans* (20%) and *T. vaginalis* (2%) in HIV-negative patients (2).

Another study indicates that there was an association of *N. gonorrhoeae*, cervicovaginal ulcer disease and cervical mucopus with cervicovaginal viral shedding of HIV-1 (26). After treatment shedding HIV-1 viral ribonucleic acid from the genital tract was reduced from 42% to 21% (26, 27).

According to WHO estimate from HIV sentinel surveillance among women attending antenatal care clinics and commercial sex workers on selected sites in Ethiopia, the prevalence of *T. pallidum* was 2.3% in antenatal care clinic attendees and 35% in commercial sex workers (28). A study from Addis Ababa showed that the prevalence of *N. gonorrhoeae* among youth was 2.9% (29).

Different literatures indicated about the emergence of drug resistance of *N. gonorrhoeae* for most of the available drugs. A study from Bangladesh showed that 66% of *N. gonorrhoeae* isolates were resistant to penicillin, 60.6% to tetracycline, 11.7% resistant and 26.6% intermediate resistant to ciprofloxacin and 1% was resistant to ceftriaxone (8).

3. SIGNIFICANCE OF THE STUDY

Sexually transmitted disease causes 10-40% of prenatal mortality in infants and labor complications for mothers and severity is higher in developing countries (9). On the other hand, most of STI management is syndrome approach. Recent evidence from a prospective study of women with symptomatic STIs in Ethiopia showed that genital ulcer disease was significantly associated with treatment failure using syndromatic management (30).

Studies on prevalence and associated risk factors of *T. vaginalis*, *N. gonorrhoeae*, *T. pallidum* and *Candida species* infections among symptomatic STI patients are generally very limited in Ethiopia in particular Gondar.

Hence, assessing the prevalence and risk factors of *T. vaginalis*, *N. gonorrhoeae*, *T. pallidum*, and *Candida species* among symptomatic STI patients will provide information for clinicians for appropriate management of STI patients. Moreover, policy makers and health professionals can use the findings to design STI related programs.

4. OBJECTIVES

4.1 General objective:

- To assess the prevalence and associated risk factors of *N. gonorrhoeae*, *T. pallidum*, *T. vaginalis* and *Candida* species infections among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

4.2 Specific objectives

- To determine prevalence of *T. pallidum*, *T. vaginalis*, *N. gonorrhoeae*, *Candida* species infections
- To identify the possible associated risk factors of identified infections.
- To determine antimicrobial susceptibility patterns of *N. gonorrhoeae* isolates.

5. MATERIALS AND METHODS

5.1. Study area

The study was conducted in Gondar town hospitals and health centers, Northwest Ethiopia. The Town is 725 km far from Addis Ababa, located in the North Gondar Zone of the Amhara Region. Gondar town is found north of Lake Tana on the Lesser Angereb River and Southwest of the Simien Mountains. The town has a latitude and longitude of 12°36'N 37°28' E / 12.6°N 37.467°E with an elevation of 2133 meters above sea level. Based on figures from 2007 Central Statistical Agency of Ethiopia, Gondar town has a total population of 207,044, of whom 98,120 are men and 108,924 women. In the town there are two hospitals (1 governmental and 1 private) and 5 health centers.

5.2. Study design and period

A Crosse -sectional study was conducted from April 1 to August 30, 2013.

5.3. Population

5.3.1. Source population

Those STI suspected patients who have access to visit Gondar town hospitals and health centers.

5.3.2. Study Population

All STI suspected patients attending Gondar town hospitals and health centers during the study period.

5.4. Exclusion criteria

Those patients under antibiotic treatment during 7 days before the enrolment of this study were excluded. Female patients who were on menstruation at the time of examination were also excluded from the study.

5.5. Variables

5.5.1. Dependent variables

- Prevalence of *N. gonorrhoeae*, *T. pallidum* and *T. vaginalis* and *Candida* species infections.
- Antimicrobial susceptibility of *N. gonorrhoeae*.

5.5.2. Independent variables

- Age,
- Sex,
- Marital status,
- Educational status
- Occupation,
- Number of sexual partners,
- History of STI,
- Sexual orientation,
- Prostitute contacts,
- History of abortion,
- Condom use
- Economical status.
- risk behaviors(alcohol, cigarette, chat consumption

5.6. Sample size and sampling technique

By using convenient sampling technique, 120 consecutive consenting patients, age of 15-45, who attended Gondar town hospitals and health centers STI clinic, with one or more of the complaints as stated by WHO in its syndromic approach for the diagnosis of STI were included (31).

5.7. Data collection Procedure

5.7.1. Socio-demographic characteristics

After taking written informed consent from each study participant, semi-structured questionnaire was used to collect socio-demographic and clinical data needed in this study. Data were collected by trained nurses.

5.7.2. Sample collection

Urethral and vaginal specimen

External inspection of the genital area were made, and the characteristics of any local changes such as; erythema, abrasions, ulceration, urethral discharge and vaginal discharge were noted including color, amount, odor and consistency. In the absence of visible urethral discharge, the

patient was asked to milk the urethra. A specimen of discharge was obtained at or just within the urethral meatus for male and both cervical and vaginal fluids were taken for female. Two cervical and one vaginal swabs were taken from females as well as three urethral swabs were taken from males; one for Gram staining, one for wet mount examination which was examined in the facilities and the other was put on Amies transport media for *N. gonorrhoeae* culture at the University of Gondar Hospital.

Venous blood collection

Five ml of venous blood, for detection *T. pallidum* and HIV was collected from the antecubital vein of each patients into sterile tubes. The blood was allowed to retract and then centrifuged, and the serum was obtained (32).

5.7.3. Laboratory methods

Isolation and Identification of *N. gonorrhea*:

Urethral swab and cervical swab specimens were inoculated onto Thayer Martin media (OXOID, UK) plates containing vancomycin, colistin, nistatin and trimetoprim and incubated under 5-10% CO₂ enriched wet atmosphere for 24-48 hours. Observing polymorphonuclear leucocytes with Gram-negative intracellular diplococci was a presumptive diagnosis of gonococcal infection. Isolates were identified as *N. gonorrhoeae* on the basis of colony morphology, Gram staining, oxidase test, and carbohydrate utilization test (32).

Antimicrobial susceptibility testing

Antimicrobial susceptibility test for *N. gonorrhoeae* to ceftriaxone 30µg, tetracycline 30µg, ciprofloxacin 5µg, clindmycin 2µg, penicillin 10µg, cefoxitin 30µg and cefotaxime 30µg (OXOID, UK) were carried out using disc diffusion method (modified Kirby-Bauer) on Thayer Martin media, following the recommended standards of the Clinical and Laboratory Standards Institutes (CLSI) (33). A 0.5 McFarland standard was used to have standardized inoculum density for susceptibility test. Within 15 minutes after adjusting the turbidity of the inoculum suspension, a sterile cotton swab was dipped into the adjusted suspension and squeezed on the wall of test tube. The dried TMT surface plates were inoculated by streaking the swab over the entire sterile agar surface. By using sterile forceps, the antimicrobial discs were placed on the lawn of bacterial isolates. Thayer Martin media was incubated in a humid, 5-10% CO₂ atmosphere for 18–22

hours at 36°C. After incubation, the diameter of each zone of inhibition was measured with a ruler from the edges of the last visible colony-forming growth by positioning the ruler across the centre of the disc. The results were recorded in millimeters and interpretation of susceptibility, susceptible, intermediate, or resistant following the CLSI guidelines (33, 34).

Wet mount preparation and Identification of *T. vaginalis*:

All discharges collected from the patients were subjected for microscopic examination to detect *T. vaginalis*. A suspension of the discharge with 0.3ml normal saline was prepared. One drop of suspension was put on a clean slide, covered with a cover slip and immediately examined microscopically under the low power (10x) and high power (40x) magnifications. *Trichomonas vaginalis* was identified by its characteristic morphology and darting motility in a wet smear (32).

Rapid immunochromatographic syphilis tests

Advanced quality one step rapid immunochromatographic syphilis tests (TPHA) (InTec PRODUCTS, INC., china) having *T. pallidum* recombinant antigens and provide results similar to those of specific treponemal tests was used. This was performed by adding 3 drop of serum sample to the sample well. Test results were read after 15 minutes. A positive test was shown by a pink-mauve line in both the control and test areas. A negative test was shown by a pink line in the control area only. The sensitivity and specificity of the test were 85–98% and 93–98% respectively (32).

Identification of *Candida* species

Candidiasis was identified in the bases of detecting yeast cells and pseudohyphae in wet vaginal/urethral preparations and large Gram positive yeast cells and pseudohyphae in Gram stained smears (32).

HIV-test

The presence of HIV-1/2 antibodies in the serum was determined using rapid HIV-1/2 diagnostic test kits according to the manufacturer's instructions. The results were then interpreted following the current national algorithm for screening of sera for HIV-1/2 infection that was adopted from WHO. In brief, the sera were first tested with KHB (Chimbo, china) HIV-1/2. If the result was negative it was taken as negative. For KHB positive results, further tests with STATPACK

(Trinity biotech PLC, Ireland) HIV-1/2 were done. If the result of STAT PACK was found positive, then the serum was considered as positive for HIV-1/2 antibodies, if not it was tested for a third time with a tiebreaker UNI-GOLD (MEDFOM, New York) (35).

5.8. Data quality control

The questionnaire was pre-tested on 15 outpatient department patients for comprehensiveness, effectiveness, reliability and validity. Training was given for data collectors on data collection procedures by support of investigator, venereologist and gynecologist. Culture media sterility was checked by incubating 5% of each batch of the prepared media at 37°C for 24 hours (33, 34). Performance of prepared media was also checked by inoculating with control strains of *N. gonorrhea* ATCC 49226.

5.9. Data analysis

Data were entered and analyzed using SPSS version 20. Descriptive statistics was applied to determine the distribution of the socio-demographic and clinical characteristics. Bivariate analysis was performed to examine possible risk factors for *N. gonorrhoeae*, *T. vaginalis*, *T. pallidum* and *Candida species* infections. To obtain adjusted estimates of the odds ratio while accounting for all confounding variables, multiple logistic regression was used. P-values < 0.05 were considered statistically significant.

5.10. Ethical considerations

The proposed study was approved by the research and ethics committee of the School of Biomedical and Laboratory Sciences of the University of Gondar. Official permission was obtained from each health facilities. The aim and details of the study was explained to each study participant before obtaining their written consent for specimen collection. Confidentiality was maintained at all levels of the study by using codes rather than names of the study participants. Participant's involvement in the study was on voluntary basis; participants unwilling to participate in the study & those who wish to quit their participation at any stage were informed to do so without any restriction. The results of all positive individuals were reported to the respective health facilities for better management of the patients.

6. RESULTS

6.1. Socio-demographic characteristics

A total of 120 symptomatic STI patients were enrolled in the study, of which 99 (82.5%) were females. The mean age of the study participants was 27.8 years with a standard deviation of ± 7.2 years. The majority of the study participants were in the age group < 30 years old (66.6%), literate (61.7%) and married (48.3%). Only 32 (26.6%) patients were employed. In terms of economical status, 37 (30.8 %) participants reported as they didn't have their own income (Table 1). All the patients come to the health facilities without their partners.

Table 1: Socio-demographic characteristics of symptomatic STI patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Variable		Frequency	Percentage
Sex	Male	21	17.5
	Female	99	82.5
Age	15-29	80	66.7
	30-45	40	33.3
Marital status	Single	49	40.8
	Married	58	48.3
	Widowed	2	1.7
	Divorced	11	9.2
Educational status	Illiterate	46	38.3
	Literate	74	61.7
Occupation	No	23	19.2
	House wife	26	21.7
	Sex worker	11	9.2
	Student	9	7.5
	Employed	42	35
	Farmer	7	5.8
	Other	2	1.6
	No	37	30.8
Monthly income	< 1200	49	40.8
	>1200	34	28.4

6.2. Prevalence of laboratory confirmed STI agents

The overall prevalence of laboratory confirmed STIs pathogens among suspected attendees of Gondar town hospitals and health centers was 89 (74.1%). The predominant infection was due to *Candida species* 39(32.5%), followed by *T. pallidum* 36(30%), *N. gonorrhoeae* 25(20.8%), *T. vaginalis* 17(14.2%). Among 36 TPHA positive cases, 8 had RPR confirmed active syphilis. Seventeen (14.2%) of the study participants were HIV positive. Majority of *N. gonorrhoeae*, *T. pallidum*, *T. vaginalis*, and *Candida species* infections were higher on aged between 15-29 years (Table 2).

Majority of the respondents (68.3%) had multiple sexual partners. The multiple sexual partners among male and female patients were 90.4% and 64.6%, respectively. From the total patients, 67 (55.8%) had history of STI and 69(57.5%) had never used condom (Table 2).

Table 2: Prevalence of laboratory confirmed STI agents among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	Total no (%)	<i>N. gonorrhea</i> no (%)	<i>T. vaginalis</i> no (%)	<i>T. pallidum</i> *** no (%)	<i>Candida</i> <i>species</i> no (%)
Sex					
Male	21(17.5)	6 (28.6)	0(0)	10(47.6)	0(0)
Female	99(82.5)	19(19.2)	17(17.2)	26(26.3)	39(39.4)
Age					
15-29	80(66.7)	17(21.3)	13(16.3)	25(31.2)	28(35)
30-45	40(33.3)	8(20)	4(10)	11(27.5)	11(27.5)
Marital status					
Single	62(51.7)	14(22.5)	12(19.3)	16(25.8)	20(32.3)
Married	58(48.3)	11(18.9)	5(8.6)	20(34.5)	19(32.7)
Educational status					
Illiterate	46(38.3)	9(19.6)	4(8.7)	17(36.9)	13(28.3)
Literate	74(61.7)	16(21.6)	13(17.5)	19(25.6)	26(35.1)
Occupation					
Employed	42(35)	11(26.1)	2(4.7)	16(38)	9(21.4)
House wife	26(21.7)	3(12)	5(29.4)	7(19.4)	9(23)
CSW	11(9.2)	2(8)	1(5.8)	6(16.6)	6(15.3)
No	23(19.2)	5(20)	3(17.6)	4(11.1)	9(23)
Others	18(15.1)	4(16)	6(33.3)	3(18.7)	6(33.3)
Income					
No	37(30.8)	7(18.9)	7(18.9)	7(18.9)	11(29.7)
<1200	49(40.8)	10(20.4)	8(16.3)	15(30.6)	12(24)
>1200	34(28.4)	8(23.5)	2(5.8)	14(41.2)	16(47.1)
Condom use					
Never	69(57.5)	13(18.8)	11(15.9)	19(27.5)	22(31.8)
Sometimes/Rarely	38(32)	9(23.3)	5(13.2)	9(23.7)	11(28.8)
Always	13(10.5)	3(23.1)	1(7.7)	8(61.5)	6(46.2)
HIV status					
Positive	17(14.2)	1(5.8)	2(11.7)	11(64.7)	6(35.3)
Negative	103(86)	24(23.3)	15(14.5)	25(24.3)	33(32)
Had a history of					
Abortion*	44(44.5)	7(15.9)	9(20.5)	16(36.4)	19(43.1)
Chew chat	18(15.0)	4(22.2)	3(16.7)	8(44.4)	4(22.2)
Drink alcohol always	26 (2.7)	8(30.8)	3(11.5)	12(46.2)	6(23.1)
Smoking	9(7.5)	3(33.3)	0(0)	5(55.5)	1(11.1)
>1 sexual partner	82(68.4)	18(21.9)	10(12.2)	27(32.9)	23(28)
New sexual contact**	42(35)	9(21.4)	4(9.5)	15(35.7)	15(35.7)
Prostitute contact+	5(23.9)	3(60)	0(0)	2(40)	0(0)
STI	67(55.5)	16(23.9)	11(16.4)	20(29.8)	22(32.8)
Share materials	39(32.5)	6(15.3)	8(20.5)	12(30.7)	14(35.9)
Overall	120 (100)	25 (20.8)	17 (14.2)	36 (30.0)	39 (32.5)

History of abortion*= females, Prostitute contact+ = males, **-last three months, ***TPHA

6.3. Prevalence of co-infections

Of the total patients, 24 (20%) had two or more pathogens. The most frequent co-infections were *T. pallidum* and *Candida species* 7 (29.2%), *T. pallidum* and *N. gonorrhoeae* 5 (20.8%) and *T. vaginalis* and *Candida species* 3 (12.5%) (Table 3). Majority of co-infections were found among house wife's 6 (25%), governmental employed 5 (20.8%) and commercial sex worker 4 (16.6%).

Table 3: Co-infection pattern of genital pathogens among symptomatic STI suspected patients attending Gondar town hospitals and health centers.

Co-Infection Number	Frequency	Percent
<i>N. gonorrhoeae</i> + <i>T. pallidum</i>	5	20.8
<i>N. gonorrhoeae</i> + <i>T. vaginalis</i>	2	8.3
<i>N. gonorrhoeae</i> + <i>Candida species</i>	2	8.3
<i>T. pallidum</i> + <i>T. vaginalis</i>	1	4.16
<i>T. pallidum</i> + <i>Candida species</i>	7	29.2
<i>T. vaginalis</i> + <i>Candida species</i>	3	12.5
<i>N. gonorrhoeae</i> + <i>T. pallidum</i> + <i>Candida species</i>	1	4.16
<i>N. gonorrhoeae</i> + <i>T. vaginalis</i> + <i>Candida species</i>	2	8.3
<i>T. pallidum</i> + <i>T. vaginalis</i> + <i>Candida species</i>	1	4.16
Total	24	100

6.4. Associated risk factors for laboratory confirmed STIs

In this study significant association was observed between HIV status and STIs. In multivariate logistic regression analysis the odds of having STI was 5.5 time higher among HIV positive patients (AOR 5.5, 95%CI 1.06-28.7, P= 0.04) compared to HIV negative patients. However, no statistical significant association was observed between STI and other study variables such as age, sex, educational status, marital status, occupation, economical status, smoking, number of sexual partner, chat chewing (Table 4).

Table 4: Bivariate and multivariate analysis of risk factors of STI among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	STI		COR (95% CI)	P	AOR (95% CI)	P
	Positive no (%)	Negative no (%)				
Sex						
Male	15(71.4)	6(28.6)	0.89(0.3-2.9)	0.82	1.1(0.41-3.2)	0.81
Female	73(73.7)	26(26.3)	1.00		1.00	
Age						
15-29	61(76.3)	19(23.7)	1.55(0.6-3.9)	0.30	0.13(0.48-4.0)	0.53
30-45	27(67.5)	13(32.5)	1.00		1.00	
Marital status						
Single	45(72.5)	17(27.5)	0.9(0.4-2.1)	0.84	1.0(0.38-2.9)	0.90
Married	43(74.1)	15(25.9)	1.00		1.00	
Condom use						
Never	55(79.7)	14(20.3)	2.46(0.6-10.2)	0.16	2.5(0.93-6.9)	0.06
Sometimes/Rarely	25(65.7)	13(34.3)	1.20(0.3-5.3)	1.00	0.75(0.17-3.2)	0.70
Always	8(61.5)	5(38.5)	1.00		1.00	
HIV status						
Positive	15(88.2)	2(11.8)	0.32(0.1-1.6)	0.23	5.5(1.06-28.7)	0.04
Negative	73(70.2)	30(29.1)	1.00		1.00	
Had a history of						
Chew chat (no=1)	16(88.9)	2(11.1)	0.30(0.0-1.5)	0.22	Undefined	0.99
Drink alcohol always(no*=1)	20(76.9)	6(23.1)	1.27(0.24-4.0)	0.64	1.5(0.43-5.1)	0.51
Smoking (no=1)	7(77.8)	2(22.2)	0.77(0.1-4.4)	1.00	Undefined	0.99
>1 sexual partner (no=1)	60(73.1)	22(26.9)	0.97(0.4-2.5)	0.95	1.0(0.36-2.7)	0.99
STI (no=1)	48(71.6)	19(28.4)	0.82(0.3-2.0)	0.64	0.75(0.30-1.8)	0.54
Share materials (no=1)	30(76.9)	9(25.1)	0.76(0.3-2.0)	0.54	0.99(0.37-2.6)	0.99

COR: crude odds ratio, AOR: adjusted odds ratio, HIV: human immunodeficiency virus, STI: sexual transmitted infection, *: rarely/sometimes, 1= reference

Higher prevalence of gonococcal infection were observed among males (28.9%), patients with history of multiple sexual partner (21.9%), history of STI (23.9%) and alcohol use (24.6%). However, in logistic regression analysis no statistically significant association was observed between the gonococcal infection and all possible associated risk factors (Table 5).

Table 5: Bivariate and multivariate analysis of risk factors of *N. gonorrhea* infection among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	<i>N. gonorrhea</i> infection		COR (95% CI)	P	AOR (95% CI)	P
	Positive no (%)	Negative no (%)				
Sex						
Male	6 (28.6)	15(71.4)	1.68 (0.5-5.5)	0.37	1.2(0.2-7.0)	0.97
Female	19(19.2)	80(80.8)	1.00		1.00	
Age						
15-29	17(21.3)	63(78.7)	1.08 (0.4-3.1)	0.87	1.4(0.4-5.0)	0.53
30-45	8(20)	32(80)	1.00		1.00	
Marital status						
Single	14(22.6)	48(77.4)	1.2(0.51-3)	0.62	0.42(0.1-1.3)	0.13
Married	11(18.9)	47(81.1)	1.00			
Condom use						
Never	18(26.1)	41(74.9)	2.41 (0.4-17.6)	0.33	3.2(0.9-11.0)	0.65
Sometimes/Rarely	5(13.5)	11(86.5)	2.50 (0.3-23.9)	0.41	0.6(0.2-11.5)	0.61
Always	2(15.4)	11(84.6)	1.00		1.00	
HIV status						
Positive	1(5.8)	16(94.1)	0.21(0.0-1.6)	0.12	0.21(0.2-1.8)	0.1
Negative	24(23.3)	79(76.7)	1.00		1.00	
Had a history of						
Chew chat (no=1)	4(22.2)	14(77.8)	1.24 (0.3-4.6)	0.75	0.2(0.0-2.9)	0.26
Drink alcohol always(no*=1)	8(30.8)	18 (69.2)	2.01 (0.7-5.9)	0.15	0.24(0.71-8.2)	0.15
Smoking (no=1)	3(33.3)	6(66.8)	2.02 (0.4-10.2)	0.39	3.6(0.2-55.0)	0.35
>1 sexual partner (no=1)	18(21.9)	64(79.1)	1.25 (0.4-3.7)	0.65	1.2(0.39-3.9)	0.71
STI (no=1)	16(23.9)	51 (76.2)	1.53 (0.6-4.2)	0.35	0.21(0.76-5.9)	0.14
Share materials (no=1)	6(15.3)	33(84.6)	0.59 (0.2-1.8)	0.31	0.54(0.7-1.6)	0.29

COR: crude odds ratio, AOR: adjusted odds ratio, HIV: human immunodeficiency virus, *: rarely/sometimes, 1= reference

T. vaginalis infection was higher among singles and age group of patients 15-19 years old. In terms of marital status, multivariate logistic regression analysis indicated that single study participants were about 6 times more likely to have *T. vaginalis* infection (AOR 5.9, 95%CI 1.18-29.9, p=0.036) as compared to married study subjects. However, there were no significant difference between *T. vaginalis* infection and other risk factors. Furthermore, all infection of *T. vaginalis* was found among females (Table 6).

Table 6: Bivariate and multivariate analysis of risk factors of *T. vaginalis* infection among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	<i>T. vaginalis</i> infection		COR (95% CI)	P	AOR (95% CI)	P
	Positive no (%)	Negative no (%)				
Sex						
Male	0(0)	21(100)	1.00	0.04	1.00	0.99
Female	17(17.2)	82(82.8)	Undefined		Undefined	
Age						
15-29	13(16.3)	67(83.7)	1.75 (0.5-6.9)	0.35	0.86(0.19-3.9)	0.85
30-45	4(10)	36(90)	1.00		1.00	
Marital status						
Single	12(19.3)	50(80.7)	2.5(0.83-7.0)	0.10	5.9(1.18-29.9)	0.03
Married	5(8.6)	53(91.4)	1.00		1.00	
Condom use						
Never	11(15.9)	58(84.1)	2.28 (0.3-51.5)	0.68	1.0(0.28-3.8)	0.95
Sometimes/Rarely	5(13.2)	33(86.8)	1.82 (0.2-45.5)		4.9(0.4-5.7)	
Always	1(7.7)	12(92.3)	1.00		1.00	
HIV status						
Positive	2(11.7)	15(88.3)	0.78 (0.1-4.2)	1.00	0.90(0.15-52)	0.91
Negative	15(14.5)	88(85.5)	1.00		1.00	
Had a history of						
Chew chat (no=1)	3(17.6)	15(82.4)	1.26 (0.3-5.5)	0.72	8.2(1.0-67.1)	0.04
Drink alcohol always(no*=1)	3(11.5)	23(88.5)	0.75 (0.2-3.1)		0.93(0.19-4.6)	
Smoking (no=1)	0(0)	9 (100)	0.00 (0.0-3.6)	0.35	Undefined	0.99
>1 sexual partner (no=1)	10(12.2)	72(87.5)	0.62 (0.2-2.0)		0.44(0.11-4.6)	
STI (no=1)	11(16.4)	56(83.6)	1.54 (0.5-5.1)	0.43	0.14(0.41-5.3)	0.54
Share materials (no=1)	8(20.5)	31(79.5)	2.06 (0.7-6.6)		1.4(0.42-4.7)	

COR: crude odds ratio, AOR: adjusted odds ratio, HIV: human immunodeficiency virus, *: rarely/sometimes, 1= reference

The prevalence of *T. pallidum* infection by TPHA was 32.5%. Of the TPHA positive patients, 6.6% of the patients were confirmed as active syphilis cases by RPR. The overall prevalence of *T. pallidum* infection was higher among HIV positive patients 64.7 % (11/17) than HIV negative 24.3 % (25/103) counterparts. In the multivariate logistic regression analysis HIV infection (OR 11.9, 95% CI 3.0-46.4, $p<0.001$) and regular alcohol intake (OR 3.9, 95% CI 1.2-12.9, $p=0.02$) were significant predictors of *T. pallidum* infection. Patients who consume alcohol in regular base were 3 times more likely to be infected with *T. pallidum* than no/non-regular alcohol users (Table 7).

Table 7: Bivariate and multivariate analysis of risk factors of *T. pallidum* infection among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	<i>T. palladium</i> infection		COR (95% CI)	P	AOR (95% CI)	P
	Positive no (%)	Negative no (%)				
Sex						
Male	10(47.6)	11(52.4)	2.55 (0.9-7.5)	0.05	0.50(0.13-1.8)	0.16
Female	26(26.3)	73(73.8)	1.00		1.00	
Age						
15-29	25(31.2)	55(68.8)	1.20 (0.5-3.0)	0.67	2.2(0.70-7.4)	0.30
30-45	11(27.5)	29(72.5)	1.00		1.00	
Marital status						
Single	14(26)	48(74)	0.6(0.3-1.4)	0.30	0.42(0.1-12)	0.13
Married	20(35)	38(65)	1.00		1.00	
Condom use						
Never	19(27.5)	50(72.5)	0.24 (0.1-0.9)	0.02	3.0(0.97-9.2)	0.55
Sometimes/Rarely	9(23.7)	29(76.3)	0.19 (0.0-0.89)	0.02	0.65(0.1-4.3)	0.66
Always	8(61.5)	5(38.5)	1.00		1.00	
HIV status						
Positive	11(64.7)	6(35.3)	5.72 (1.7-19.7)	0.00	11.9(3.0-46.4)	0.00
Negative	25(24.3)	78(75.7)	1.00		1.00	
Had a history of						
Chew chat (no=1)	8(44.5)	10(55.5)	2.11(0.7-6.6)	0.15	0.46(0.6-3.2)	0.43
Drink alcohol always(no*=1)	12(46.1)	14(53.9)	2.50 (0.9-6.8)	0.04	3.9(1.2-12.9)	0.02
Smoking (no=1)	5(55.5)	4(44.5)	3.23 (0.7-15.6)	0.13	2.3(0.23-23)	0.46
>1 sexual partner (no=1)	27(32.9)	55(67.1)	1.58 (0.6-4.2)	0.30	1.4(0.48-4.2)	0.52
STI (no=1)	20(29.8)	47(70.2)	0.98 (0.4-2.3)	0.96	0.76(0.29-1.9)	0.58
Share materials (no=1)	12(30.7)	27(69.3)	1.06 (0.4-2.6)	0.89	0.87(0.31-2.4)	0.29

COR: crude odds ratio, AOR: adjusted odds ratio, HIV: human immunodeficiency virus, *: rarely/sometimes, 1= reference

All the *Candida* species were isolated from female patients only. The prevalence of *Candida* species was higher in HIV positive patients 35.3% (6/17) than HIV negative patients 32% (33/103). However, none of the study variables were found to be significantly associated with the prevalence of *Candida* species ($P > 0.05$) (Table 8).

Table 8: Bivariate and multivariate analysis of risk factors of *Candida* species infection among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Characteristics	<i>Candida</i> species infection		COR (95% CI)	P	AOR (95% CI)	P
	Positive no (%)	Negative no (%)				
Sex						
Male	0(0)	21(100)	1.00	0.00	1.00	0.99
Female	39(39.4)	60(60.6)	Undefined		Undefined	
Age						
15-29	28(35)	52(65)	1.42 (0.6-3.6)	0.40	1.4(0.49-4.2)	0.49
30-45	11(27.5)	29(72.5)	1.00		1.00	
Marital status						
Single	20(32.5)	42(67.5)	0.97(0.45-2)	0.95	1.00	0.98
Married	19(32.7)	39(67.3)	1.00		1.0(0.36-2.8)	
Condom use						
Never	25(36.3)	44(63.7)	0.91 (0.2-3.6)	1.00	1.5(0.57-4.1)	0.39
Sometimes/Rarely	9(23.7)	29(76.3)	0.50 (0.1-2.3)	0.31	1.6(0.38-7.3)	0.49
Always	5(38.5)	8(61.5)	1.00		1.00	
HIV status						
Positive	6(35.3)	11(64.7)	1.16 (0.3-3.8)	0.79	1.6(0.45-5.7)	0.46
Negative	33(32)	70(68)	1.00		1.00	
Had a history of						
Chew chat (no=1)	4(22.2)	14(77.8)	0.55 (0.1-1.9)	0.31	1.1(0.19-6.4)	0.90
Drink alcohol always(no*=1)	6 (23)	20(77)	0.55 (0.2-1.7)	0.25	1.0(0.30-3.5)	0.95
Smoking (no=1)	1(11.1)	8(88.9)	0.24 (0.0-2.0)	0.27	0.77(0.44-14)	0.86
>1 sexual partner (no=1)	23(28)	69(72)	0.46 (0.2-1.1)	0.05	0.74(0.29-1.8)	0.53
STI (no=1)	22(32.8)	45(67.2)	1.04 (0.5-2.4)	0.93	0.97(0.39-2.4)	0.95
Share materials (no=1)	14(35.9)	25(64.1)	1.32 (0.6-3.2)	0.50	0.87(0.35-2.1)	0.77

COR: crude odds ratio, AOR: adjusted odds ratio, HIV: human immunodeficiency virus, *: rarely/sometimes, 1= reference

6.5. Antimicrobial susceptibility pattern of *N. gonorrhea* strains

The antimicrobial susceptibility pattern of all isolates of *N. gonorrhea* is shown in Table 9. All isolated *N. gonorrhea* showed resistance to at least one antimicrobial agent. The susceptibility pattern of isolates showed that there were 100% non-susceptibility to tetracycline, 76% non susceptibility and 24% intermediate resistant to penicillin. Of isolated *N. gonorrhea*, 52% were resistant to ciprofloxacin, 48% to ceftriaxone, 44% to cefoxitin, 32% to cefotaxime and 28% to clindmycin. Most of the isolates (80%) were shown multiple drug resistance and 11(44%) of the isolates were non-susceptible to both ciprofloxacin and ceftriaxone.

Table 9. Antimicrobial susceptibility pattern of *N. gonorrhea* isolated (n=25) among symptomatic STI suspected patients attending Gondar town hospitals and health centers from April 1 to August 30, 2013.

Antibiotic	S	I	R
Penicillin	0(0)	6(24)	19(76)
Tetracycline	0(0)	0(0)	25(100)
Ciprofloxacin	4(16)	8(32)	13(52)
Ceftriaxone	13(52)	-	12(48)
Cefotaxime	17(68)	-	7(28)
Cefoxitin	14(56)	-	11(44)
Clindmycin	18(72)	-	7(28)

R; resistance, I; intermediate, S; sensitive,

7. DISCUSSION

Data are scarce regarding the epidemiology of STIs in most countries due to various reasons such as stigma and discrimination associated with the STI, poor attendance of STI patients at the public health facilities and scarcity of diagnostic facilities. The situation induces many developing countries, including Ethiopia, to corporate syndromic approach for STI diagnosis and treatment (10).

The overall prevalence of laboratory confirmed STI excluding HIV was 74.1%. This result was consistent with a study done almost two decades ago in Egypt (71.6%) (36). A higher prevalence (85%) was found, in reports from Nigeria a decade ago (37). However, comparatively lower prevalence (39%) was reported in Ibadan, Nigeria (21). The difference in prevalence rates might be due to the difference in geographical location and laboratory diagnostic methods used.

In this study prevalence of *N. gonorrhoeae* infection (20.8%) was comparable with studies from Mozambique (22.5%) (38), Egypt (26%) (36) and Southwestern Nigeria (25%) (23). However, it was higher than the reports from different countries such as Vientiane (0.8%) (39), Mongolia (11%) (17), Colombia (1.4%) (1), India (6%) (20) and Nigeria (1%) (40). Moreover, in our study the prevalence of *N. gonorrhoeae* infection was four fold higher than reported by Hailemariam *et al.* from Hawassa, Ethiopia (5.1%) (41). This discrepancies in the prevalence of *N. gonorrhoeae* might be due to socioeconomic, cultural, geographic, development of resistance for drugs used on syndromic management, lack of differential diagnosis and poor partner tracing system specially in our settings.

In this study the prevalence of *T. pallidum* infection by TPHA was 32.5%. Among this, 6.6% of the patients were confirmed as active syphilis cases by RPR. The prevalence of active syphilis in this study was comparable to report from India (6%) (20). This finding was higher than the reports from Nigeria (1.5%) (39) and 0% prevalence among ANC clients, southern Ethiopia (42). However, our finding was relatively lower as compared to 8% in rural South Africa (43) and 15% in rural Mozambique (44). This may be due to the difference in study population and syphilis can effectively treated with single dose of bezantine penicillin. Thus the prevalence declines with time.

In our study, the prevalence of *T. vaginalis* was 14.5%. This finding was comparable with the report from South Africa in which the prevalence of *T. vaginalis* among family planning service user was 14% (43). However, it was higher than the reports from Maputo and Ibadan where the prevalence of *T. vaginalis* was 2% for each (38, 40). Furthermore, lower prevalence of *T. vaginalis* were reported from HIV positive and HIV negatives women attending HIV clinic (12%) and STI clinic (2%) in Nigeria (2), and 1.5% in Egypt (36). In the contrary, our finding was lower than the prevalence of *T. vaginalis* (24.4%) among HIV-seropositive Nigerian women (45). These differences may be due to variation in time and among study population.

In our study the prevalence of *Candida species* infection (32.5%) was highest as compared to the prevalence of *N. gonorrhoeae*, *T. pallidum* and *T. vaginalis* infections. This prevalence of *Candida species* was lower as compared to the report from Nigeria (39%) (2). However, lower findings were reported from Colombia (11%), Nigeria (27%), Tanzania (25.5%), and Ethiopia (16%) (1, 21, 46,47).

Risk factors associated with STI in this study was observed between positive HIV status and STI. In a multivariate analysis the odds of having STI was 5 fold higher among HIV positives than their counter parts. Keeping the finding reported from Nigeria (2) HIV infections was significantly associated with STI among symptomatic patients. Moreover, the prevalence rate of STIs was higher (44%) among age group of 15-24 years old as compared to the other age groups. This observation had in conformity with generally observed fact that the incidence of STIs by the number of cases treated each year is highest among 15-24 years old (48).

Although high gonococcal infection were observed among male patients (29%), clients having multiple sexual partner (22%), patients with history of STI (23.9%) and alcohol use (24.6%), there were no statistically significant association observed between the gonococcal infection and all possible associated risk factors. In contrast to our finding, reports from Ibadan, Southwestern Nigeria (21), (22) and north-west Tanzania (46) indicated that marital status, sex and HIV status were significant predictors of *N. gonorrhoeae* infections.

In this study, the prevalence of *T. pallidum* was higher among HIV positive patients (64.7 %) as compared to HIV negative (24.3 %) counterparts. Furthermore, logistic regression analysis indicated that HIV infection was significant predictor of *T. pallidum*. This finding was consistent

with reports from Kenya (49) and Addis Ababa, Ethiopia (50). Regular alcohol intake also had significant association with *T. pallidum* infection (OR 3.9, 95% CI 1.2-12.9, p=0.032). This is comparable to study from Colombia (1) and among female commercial sex workers Addis Ababa, Ethiopia (51).

Keeping the report from Nigeria (21), in this study being single was significant risk factor for *T. vaginalis* infection. Females had highest prevalence of *T. vaginalis* compared to male patients. This finding is comparable to the study conducted in Nigeria (39). In contrast to the reports from Nigeria (21) and Australia (52) in our study the prevalence of *T. vaginalis* was higher among age group of 15 - 19 years old patients (54.4%) and prevalence decreases when age increases. This higher prevalence of *T. vaginalis* infection among young age group may be due to poor sanitation and knowledge about STI.

Generic WHO guideline recommended that countries with resource poor settings should use syndromic approach for STI management though in recent years there has been increased antimicrobial resistance of *N. gonorrhoeae* isolates in different parts of the developing countries. Regimens for treatment of STI range from oral or injectable fluoroquinolones and cephalosporins.

In this study High levels of resistance to tetracycline and penicillin observed have agreement with report from Malawi (53). A comparable results of tetracycline (100%) and penicillin (100%) resistant *N. gonorrhoeae* isolates among gynecological patients were reported from Hawassa, Ethiopia (41). Similarly high prevalence of antimicrobial resistant *N. gonorrhoeae* isolates against tetracycline (92.6%) and penicillin (94.4%) were reported from northwest Ethiopia (54).

Currently Ethiopian ministry of health recommends single dose 500mg ciprofloxacin or 1g ceftriaxone as first drug of choices for treatment of gonorrhea (55). In this study high prevalence drug resistant *N. gonorrhoeae* isolates to ciprofloxacin (52%) and ceftriaxone (48%) were observed. Concordant findings in the prevalence rate of ciprofloxacin resistant *N. gonorrhoeae* were reported from Northwest Ethiopia (40.9%) (54) and Tanzania (77%) (46). In contrast to our finding, study reported from Hawassa, Ethiopia, indicated that all isolated *N. gonorrhoeae*

were susceptible to ceftriaxone however, 18% of the isolates were resistance to ciprofloxacin (41). Similarly Tibebu *et al* reported 40.9% and 27.8% non susceptibility of *N. gonorrhoeae* to ciprofloxacin and ceftriaxone respectively (54). The discrepancy in *N. gonorrhoeae* drug susceptibility pattern may be the difference in use of the two drugs for the treatment of other diseases and the time variation.

Furthermore study conducted in Bangladesh among commercial sex workers, prevalence of ceftriaxone resistant gonococci strains were 1% (8). In this study resistance rate of *N. gonorrhoeae* isolates were relatively lower against cefotaxime (28%) as compared to ceftriaxone (48%), though it was lower as compared to study finding reported from Hawassa (41). Similar to cefotaxime, the lower resistance rate of isolated *N. gonorrhoeae* (28%) to clindamycin which is mostly prescribed drug for pelvic inflammatory disease was observed. Even though the result is not shown, *N. gonorrhoeae* isolates shows high non-susceptibility for erythromycin and doxycycline which are use as supplement treatments for uncomplicated *gonorrhoeae*. The higher resistance rate *N. gonorrhoeae* isolates observed in this study may be due to the use of gonococcal drugs for the treatment of other bacterial diseases, the presence of poor illegal drug consumption, and lack of etiologic management.

8. LIMITATION OF THE STUDY

As the study was conducted on small number of STI suspected symptomatic patients, it may not show the epidemiology and determinants of these STI causing pathogens in the community. All drugs used for the treatment of gonococcal infections were not tested.

9. CONCLUSION

Higher prevalence of *N. gonorrhoeae*, *T. pallidum*, *T. vaginalis*, and *Candida species* infections among STI suspected patients attending Gondar town hospitals and health centers were observed. The prevalence of *Candida* species and *T. palladium* were higher in HIV positive patients. Unmarried patients were at higher risk of *T. vaginalis* infection. Being HIV positive and regular alcohol intake were significant predictors of *T. pallidum* infection. Isolated *N. gonorrhoeae* showed higher antimicrobial resistance to penicillin, tetracycline, ciprofloxacin and ceftriaxone.

10. RECOMMENDATIONS

For better understanding of the epidemiology of STI causing pathogens and the association between different patient variables and STI associated infectious diseases large scale community based study should be considered. Emphasize on partners tracking system (partner notification or patient-delivered partner treatment) and education about proper condom use as one of prevention and controlling strategy of STIs. Microbiological diagnosis and susceptibility testing of gonococcal infections should be strengthened for early detection and appropriate management of gonorrhea. Moreover, rational use of antibiotics and continuous antimicrobial surveillance should be practiced in order to tackle spread of drug resistance *N. gonorrhea* infections.

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12. ANNEXES

Annex I. English version study participant information and consent form

Consent Form for Participation in Research Work

Date: _____ Study Number: _____

Title: Prevalence and associated risk factors of *Neisseria gonorrhoeae*, *Treponema palladium* *Trichomonas vaginalis* and *Candida* species among symptomatic STI suspected patients attending Gondar town hospitals and health centers, Gondar, Northwest Ethiopia.

Purpose: Study to Determine and associated risk factors of *Neisseria gonorrhoeae*, *Treponema palladium*, *Trichomonas vaginalis* and *Candida species* among patients attending Gondar town hospitals and health centers.

Procedure to be carried out: We kindly invite you to take part in our project that 120 clients will participate. If you are willing to join in our project we are so happy for participate in this study and we need you to clearly understand the aim of this study and to sign the consent form.

As agreed to take part, you will be asked some questions relevant to the study using questionnaire. A blood sample and urethral or vaginal swab will be collected. from sample etiology of Gonorrhea, Trichomoniasis and syphilis will be examined.

Risk associated with the study: By participating in this research project you may feel that it has some discomfort during giving blood and vaginal/urethral swab. But this may not be too much as you are one of the member of the community who are at risk of this public health problem. So your response will help as an important input to show extent of the stated problem and help to design appropriate strategies to manage it. There is no risk in participating in this research project.

Benefits of this study: If you are afflicted with STI, You will be treated.

Confidentiality of your information: All information gathered from the study will remain confidential. Your participation in this study is strictly anonymous.

Right to Refusal or Withdraw: You have the full right to refuse from participating in this research and this will not affect you from getting any kind healthcare service. You have also the full right to withdraw from this study at any time you wish, without losing any of your right.

Person to contact: If you have any problems or questions related to this study you can contact telephone to Investigator

Name : Rozina Ambachew,

Telephone number 0920130978/0935706022.

email: nanieyasu@gmail.com

Informed consent form: You are invited to participate in a study on Prevalence and associated risk factors of *Neisseria gonorrhoeae*, *Treponema palladium*, *Trichomonas Vaginalis* and *Candida species* among symptomatic STI patients attending Gondar town hospitals and health centers. If you have fully understood what will be done in the course of this study and agree to the procedure involved, please sign the consent form.

I, the undersigned, confirm that the study was explained to me in a language that I understand. The objectives, risks and benefits of the study were clearly explained to me. I am also assured that participation in the study is voluntary.

Signature : _____ Date : _____

Annex II - English version Questionnaire

Name of health facility _____

Date of Interview: ____/____/____

Name of Interviewer: _____

Respondent ID Number _____

Facility ID _____

Age _____ (in complete years).

Sex Male ☐ Female ☐

Marital status: Single ☐ Married ☐ Divorced ☐ Widowed ☐

Educational background

Illiterate ☐ read and write only ☐ Primary school ☐

Secondary school ☐ College/university ☐.

5. Occupation

6. Do you drink alcohol?

Yes ☐ No ☐

6.1 If yes how often do you use?

Always ☐ Sometimes ☐ Never ☐

7. Do you chew chat?

Always ☐ Sometimes ☐ Never ☐

8. Do you smoke cigarette and/or other smokes ?

Always ☐ Sometimes ☐ Never ☐

9. Have you ever had Abortion (for female)

Yes ☐ No ☐

9.1 if yes how many times ?

10. How many sexual partners do you have in your life time?

Did not have ☐ 1 ☐ 2 ☐ ≥ 3 ☐

10.1 If yes, did you use condoms?

Always ☐ Sometimes ☐ Never

11. Have you ever had sex with prostitutes?

Yes No

11.1 if yes how many times _____

11.2 did you use condoms?

Yes No

12. Have you ever been treated for any sexually transmitted disease within last 12 months?

Yes No

17. Have you ever share any underwear, trauther, and toilet bottling before?

Yes ☐ No ☐

18. Have you ever had a blood test for HIV

Yes No Don't know

If yes, what was the result?

Positive (infected) Negative (not infected) Don't know

Annex III. Amharic version study participant information and consent form
የምርምር ተሳታፊዎች ጥናት ማብራሪያ ቅፅ

ቀን-----

መለያ ኮድ-----

የተመራማሪው ስም:- ሮዚና አምባቸው

የጥናቱ ርዕስ:- የጨብጥ፣ የቂጥኝና የቲባጅናሊስን ስርጭትና ለስርጭታቸው አስተዋጾ የሚያደርጉ ምክኒያቶችን ማወቅ።

የጥናቱ አላማና ጥቅም:- በጎንደር ከተማ በሚገኙ ጤና ተቋማት ላይ ባሉ የአባላዘር በሽታ ህክምና መስጫ ክፍሎች ላይ የሚመጡ ህመማን ያለውን የአባላዘር በሽታ ስርጭት እና ለስርጭቱ አስተዋጾ የሚያበረክቱ ምክኒያቶችን ማወቅ እንዲሁም የጨብጥ በሽታ አሁን ለመንጠቀምባቸው

የስራው ቅደም ተከተል:- በጥናቱ ውስጥ 120 ሰዎች ይሳተፋሉ። በዚህ ጊዜ ውስጥ ስላለውት የጾታዊ ባህሪ እንዲሁም ስለ አባላዘር በሽታ ስሜቶች እና ምልክቶች እንጠይቅዎታለን።

5 ሚሊ ሊትር ደም እና የብልት ፈሳሽ ይወስድለዎታል። ከተወሰደው ናሙና ላይም ቂጥኝ፣ ጨብጥና ቫጅናሊሲ ይሰራል። ውጤቱን ካርደውን በማሳየት ይወስዳሉ። ውጤትዎ ሚስጥሩ የተጠበቀ ነው ።

በዚህ ጥናት ውስጥ በመሳተፎዎ ሊከሰቱ የሚችሉ ስጋቶችን የምቶት መጓደሎች ለጥናቱ በሚወሰደው ናሙና ምክንያት ሊከሰት የሚችል የተለየ ችግር እና የሚያስጋ ምንም አይነት ነገር የለም ። ነገር ግን ናሙናው በሚወሰድበት ጊዜ መጠነኛ የሆነ የህመም ስሜት ሊያስከትል ይችላል ይህ የህመም ስሜት ምንም አይነት ችግር አያመጣም።

ከጥናቱ የሚገኘው ጥቅም:- ተሳታፊዎች በገፅ የላቭራቶሪ ውጤቱን ያገኛሉ

የሚስጥር አጠባበቅ:- ጥናቱ ላይ የሰጡት ማንኛውም መረኛ የሚቀመጠው በሚስጥር ነው። ማንኛውም ሰው ስለሰው ውጤት የሚያውቀው ነገር ስለማይኖር ስጋት አይገባዎትም።

በጥናቱ መሳተፍ:- በዚህ ጥናት መሳተፍ በፍቃደኝነት የተመሰረተ ነው።

የተመራማሪው ኢ-ሜል nanieyasu@gmail.com ስልክ 0920130978/0935706022

Annex V . Laboratory (Culture) reporting Format

D a t e	cod e	Card no	TV	Candi d specie s	Gram negative diplococci		Growth on TMT	Ox.	Cat	Super oxidase	Sensiti vity			BC			
					Intra	Extra					S	I	R	G	G	L	M

Key: Y= Yes, N=No, **Key: TV= *T. vaginalis*** TE=tetracycline, CIP= ciprofloxacin, CRO= ceftriaxone, CTX= cefotaxime, P= penicillin, FOX=cefoxitin, CLD= clindmycin, OX= oxidase, CAT= catalyse, S=Sensitive, I=Intermediate, R=Resistance, Intra= Intracellular, Extra= Extracellular, BC=Biochemical Tests

DECLARATION

I, the undersigned, infectious and tropical diseases student declare that this thesis proposal work is my original work in partial fulfillment of the requirement for the degree of master in infectious and tropical diseases.

Name: Rozina Ambachew

Signature: -----

Place of submission: School of Biomedical and Laboratory science, College of Medicine and Health science, University of Gondar

Date of submission-----

This thesis work has been submitted for examination with my/ our approval as university advisor(s)

Advisors

Name	Signature	Date
1. Mr. Abate Assefa	_____	_____
2. Dr. Beyene Moges	_____	_____
3. Mr. Mengistu Endris	_____	_____

Examiner Name	Signature
1. -----	-----
2. -----	-----

ASSURANCE OF INVESTIGATOR

The undersigned agrees to accept responsibility for the scientific, ethical and technical conduct of the research project and for provision of required progress reports as pre terms and conditions of the research and publications office of the University of Gondar.

Name of the student: Rozina Ambachew .

Date: _____ .Signature: _____ .

Approval of the advisors

Advisors

Name	Signature	Date
1. Mr. Abate Assefa	_____	_____
2. Dr. Beyene Moges	_____	_____
3. Mr. Mengistu Endris	_____	_____